

Table 2. Average percent cumulative mortality and percent inhibition of emergence (EI) of *Anopheles pharoensis* larval and pupal isolates from Dimilin-treated ditches (15.2 AI/acre) in Gezira, Sudan.

| Ditch | Larval isolates* | | | | Pupal isolates* | | |
|-------------|------------------|-------|--------|-------|-----------------|--------|-------|
| | Larvae | Pupae | Adults | (EI%) | Pupae | Adults | (EI%) |
| 1 | 20 | 0 | 0 | 100 | 20 | 2 | 90 |
| 2 | 20 | 2 | 1 | 95 | 20 | 3 | 85 |
| 3 (control) | 20 | 19 | 17 | 15 | 20 | 18 | 10 |

* Based upon the average of 6 isolates, each containing 20 larvae or pupae in a cup.

Mortality of the surviving larvae and pupae isolated from the field was high. Additional mortality occurred in subsequent stages producing a very high overall percent emergence inhibition (Table 2). Similar findings were reported by Mulla et al. (1974), Mulla and Darwazeh (1975) and Schaefer et al. (1975) using Dimilin against various mosquito species.

Thus, Dimilin offers good potential for the control of various species of mosquitoes, showing a varying degree of efficacy against different species breeding in diverse habits. This compound exhibits little or no hazard to nontarget organisms in mosquito breeding sources (Miura and Takahashi 1975, Mulla et al. 1975, Steelman et al. 1975) and has the potential for rendering control of immature mosquitoes for 10 days at very low practical rates.

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THE USE OF MEPACRINE HYDROCHLORIDE TO CONTROL *VORTICELLA* ON MOSQUITO LARVAE

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One of our recent projects entailed the recovery of eggs of the flood-water mosquito *Aedes juppi* McIntosh from soil samples collected in the Orange Free State province, South Africa, and their subsequent rearing to adults. However, only 34% of the viable eggs which hatched were reared successfully to adulthood owing to mortality of the 3rd and 4th instar larvae. Examination of dying and recently dead larvae revealed a severe infestation with the ciliate *Vorticella* which appeared to be the main cause of death. No fungi or other parasitic Protozoa were seen at this time on the larvae and the possible existence of pathogenic bacteria or viruses was not investigated. It was therefore decided to carry out a series of experiments to find a chemical which could be incorporated in the water in the larval rearing trays to kill off the *Vorticella* encrustation without harming the larvae themselves. Metronidazole and chloroquine were each

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tried unsuccessfully but another chemical, the anti-malarial mepacrine hydrochloride 4-(3-Chloro-7-methoxy acridin-9-ylamino)-NN-diethyl-pentylamine dihydrochloride dihydrate, gave promising results. It was thought likely that this would kill ciliates as it is known to kill both sporozoan and flagellate Protozoa (Bowman and Rand 1980). This article describes the results of these tests.

Mepacrine hydrochloride was dissolved in distilled water so as to provide a series of 5 concentrations from 0.0009 to 0.0045%. Infested 3rd or 4th instar larvae of *Ae. juppi*, recently dead, were placed into each of the 5 solutions. The lowest concentration at which all the *Vorticella* died was 0.0018% and the time required 21 hr. The discovery of *Vorticella* on larvae of *Ae. juppi* prompted us to examine our healthy colonies of *Culex perexiguus* Theobald and *Culex univittatus* Theobald and *Vorticella* was found on dead larvae but not on any living larvae of the 2 species. The vorticellids on these 2 mosquitoes appeared to differ morphologically and that infesting *Cx. perexiguus* also differed from the vorticellid infesting *Ae. juppi*. This may explain why less than 4 hr exposure to 0.0018% mepacrine hydrochloride was needed to kill the *Vorticella* on *Cx. perexiguus* larvae but 21.5 hr for the *Vorticella* species on *Cx. univittatus* larvae. This suggests that the *Vorticella* species on *Cx. univittatus* may have been the same as that infesting *Ae. juppi*. When dead infested *Ae. juppi* larvae were mixed with live uninfested *Cx. univittatus* 2nd instar larvae and cultured together for 2 weeks, the *Vorticella* did not spread to the *Cx. univittatus* larvae while they were alive. However, if a *Cx. univittatus* larva died from another cause, after death it swiftly became infested with *Vorticella*.

Having established the minimum concentration of mepacrine hydrochloride and the length of exposure to kill the *Vorticella*, experiments were undertaken to ascertain whether the same concentration would be detrimental to the larva itself. Since at the time of the tests no live *Ae. juppi* were available, experiments were performed using other species of mosquitoes from laboratory colonies. Exposure of mature larvae of *Cx. univittatus* for up to 8 days to mepacrine hydrochloride at concentrations greater than 0.0018% caused a 54% mortality mainly in the subsequent pupal and adult stages. The mortality rate at 0.0018% was not established although it was lower than 50%. When eggs of *Aedes aegypti formosus* (Walker) were hatched using the technique of Novak and Shroyer (1978), except that the hatching tubes contained 0.1% nutrient broth made up in 0.0018% mepacrine hydrochloride, 3 days later 10–85% of the young larvae were

found dead. When the rearing of the surviving larvae was continued in dishes of mepacrine hydrochloride, 6–25% of them failed to become adults. If eggs of *Ae. aegypti formosus* were hatched in the same way but the young larvae were transferred on the first and second days to distilled water and fed in the usual way, about 87% of these larvae reached adulthood.

In conclusion, there are two possible procedures for using mepacrine hydrochloride to control *Vorticella* infestations. In the first, eggs are hatched in 0.0018% mepacrine hydrochloride made up in 0.1% nutrient broth and the first instar larvae are transferred to clean water a minimum of 21 hr later. The second procedure is to place 2nd, 3rd and 4th instar infested larvae into mepacrine hydrochloride solution for at least 21 hr and afterwards transfer them to clean water in rearing dishes.

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EVALUATION OF A SUSTAINED RELEASE FORMULATION OF *BACILLUS THURINGIENSIS* (H-14) FOR CONTROL OF WOODLAND *CULEX* MOSQUITOES¹

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Bacillus thuringiensis var. *israelensis* (B.t.i.), or *B. thuringiensis* (H-14) as designated by de Barjac (1978), is a bacterial agent effective against several nematoceros dipteran families, including mosquitoes (Culicidae) and blackflies (Simuliidae). During sporogenesis of the bacterium, a proteinaceous crystal or parasporal body is formed. This crystal contains a delta-endotoxin which is responsible for most of the

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